## Figure 1: Top 10 BLASTP hits for INSP179 polypeptide sequence (SEQ ID NO:10) against NCBI-nr

BLASTP 2.2.2 [Jan-08-2002]

Reference: Altschul, Stephen F., Thomas L. Madden, Alejandro A. Schaffer, Jinghui Zhang, Zheng Zhang, Webb Miller, and David J. Lipman (1997), "Gapped BLAST and PSI-BLAST: a new generation of protein database search programs", Nucleic Acids Res. 25:3389-3402.

Query= INSP179.pp (880 letters)

Database: All non-redundant GenBank CDS translations+PDB+SwissProt+PIR+PRF

1,594,288 sequences; 522,190,286 total letters

Searching......done

| Sequences producing significant alignments:                         | Score<br>(bits) | E<br>Value |
|---|-----------------|------------|
| ref XP_291099.2  similar to hypothetical protein MGC38937 [Homo     | 1747            | 0.0        |
| ref NP 705796.1  hypothetical protein MGC38937 [Mus musculus] >g    | . 684           | 0.0        |
| ref   XP 223356.1   similar to hypothetical protein MGC38937 [Rattu | 650             | 0.0        |
| ref[XP 221358.2] similar to Carboxypeptidase N 83 kDa chain (Car    |                 | 5e-14      |
| ref[NP 570843.1] leucine rich repeat containing 15; leucine-rich    |                 | 2e-12      |
| gb AAH25836.1  1300018K11Rik protein [Mus musculus]                 |                 | 7e-12      |
| ref[XP 148373.1] RIKEN cDNA 1300018K11 [Mus musculus] >gi 380806    | . 75            | 7e-12      |
| ref[NP 659551.1] leucine-rich repeat protein induced by beta-amy    | . 74            | 1e-11      |
| dbj BAD01045.1  toll-like receptor 3 [Paralichthys olivaceus]       | 73              | 3e-11      |
| dbj BAB85498.1  18 wheeler [Bombyx mori]                            | 72              | 5e-11      |

## Figure 2: Pairwise alignment of INSP179 to top annotated BLASTP hit (mouse ortholog)

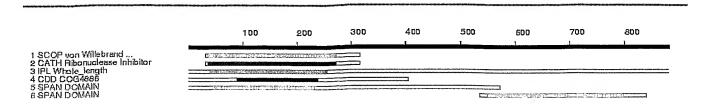
Score = 684 bits (1765), Expect = 0.0 Identities = 422/888 (47%), Positives = 529/888 (59%), Gaps = 41/888 (4%) MKNLYFRVITIVIGLYFTGIMTNASRKSNILFNSECQWNEYILTNCSFTGKCDIPVDISQ 60 Query: 1 M++ Y RV +V GL F +T SRKS++ FN E Q N +L N S MRDFYVRVTILVTGLCFVETVTTPSRKSSVSFNPEYQRNGDLLVNWSSIRH-----VSQ 54 Sbjct: 1 Query: 61 TAATVDVSFNFFRVLLQSHTKKEEWKIKHLDLSNNLISKITLSPFAYLHALEVLNLSNNA 120 +D SF FFRVL Q HT+KE IK D +++ ISK+TL P A+LHALE+LNLSN A Sbjct: 55 NTDAMDRSFYFFRVLFQPHTQKER-HIKPPDRTHHRISKVTLDPLAHLHALEILNLSNKA 113 Query: 121 IHSLSLDLLSPKSSWVKRHRSSFRNRFPLLKVLILQRNKLSDTPKGLWKLKSLQSLDLSF 180 IH SLD P SS KRH +R P L+VLILQRN+LS TPKGLWKLKSL+SLDLSF Sbjct: 114 IHYFSLDQPLPPSSHQKRHGGHSHSRLPRLQVLILQRNQLSGTPKGLWKLKSLRSLDLSF 173 Query: 181 NGILQIGWSDFHNCLQLENLCLKSNKIFKIPPQAFKDLKKLQVIDLSNNALITILPMMII 240 N I+ IG SDFH CLQLE++ LKSNKI I P+AFK LKKLQV+DL +NAL T++P++ I Sbjct: 174 NRIVHIGLSDFHGCLQLESIYLKSNKICTIHPKAFKGLKKLQVVDLRSNALTTLVPIVTI 233 Query: 241 ALEFPHLVVDLADNNWQCDDSVAVFQNFISESWRKKWNVICNRSIGSEEANGGTPQSRIS 300 ALE PHL + LADN WQC +S FQN S SWR+ W ICN S+ ++ N T Q R S Sbjct: 234 ALELPHLELGLADNQWQCSESNVNFQNITSSSWREIWKAICNMSVENKRPNAETHQIRKS 293 Query: 301 RETRL---PPIHLHRMKSLIRSKAERPQGGRHTGISTLGKKAKAGSG-LRKKQRRLPRSV 356 R+T L PP L KSLI+SKAERPQ G +S LGK+AK G G LR + P + Sbjct: 294 RDTHLLLSPPSDL---KSLIQSKAERPQAGMDMHLSALGKEAKDGYGDLRGMWPQSPVEL 350 Query: 357 RSTRDVQAAGKKEDAPQDLALAVCLSVFITFLVAFSLGAFTRPYVDRLWQKKCQSKSPGL 416 R ++D Q +K+D P L LA+CLSVFITF+VAF LGAF RPY+DRL Q++C +K PG Sbjct: 351 RDSQDEQVTDRKDDKPPALELAICLSVFITFVVAFCLGAFARPYIDRLRQQRCSNKRPGS 410 Query: 417 DNAYSNEGFYDDMEAAGHTPHPETHLRQVFPHLSLYENQTPFWVTQPHPHATVIPDRTLG 476 DNAYSN+GF+ D+E A H + T L Q HL L ENQ P WV +P PH+ V ++ LG Sbjct: 411 DNAYSNKGFHGDIEGAQHMEYQGTDLHQTTHHLHLSENQNPSWVAEPIPHSAVQSEQMLG 470 Query: 477 RSRKDPGSSQSPGQCGDNTGAGSGND-----GAVYSILQRHPHAGNRELMSAAQDHIHR 530 + DPG QSP Q D+ + SG+ + L P+A + +S Q H Sbjct: 471 SNGTDPGHQQSPEQLKDSNESRSGDSIVLPSGPVAHLALHGLPNADAHKAISPVQ---HH 527 Query: 531 NDILGEWTYETVAQEEPLSAHSVGVSSVAGTSHAVSGSSRYDSNELDPSLSGEITASLCK 590 +D L E Y+TVAQE L + SS+ G S ++L PS ++ AS K Sbict: 528 HDFLEEAHYDTVAQEYSLIDDVMDRSSITGPLGTFPSSVESRRDDLHPSQPRDVVASFSK 587 Query: 591 MLTHAEAQRTGDSKERGGTEQ-SLWDSQMEFSKERQVSSSIDLLSIQQPRLSGARAEEAL 649 L HA + S E G E DSQM S+ERQVS+SI L+ QQP G AEE L Sbjct: 588 TLAHANTREAEGSMETGCPEPLGAMDSQMGSSEERQVSNSIRELATQQPSFQGVDAEERL 647 Query: 650 SAHYSEVPYGDPRDTGPSVFPPRWDSGLDVTPANKEPVQKSTPSDTCCELES--DCDSDE 707 S YSEV + DP PS+ PRW SG V PA EPV++ P D +L + + DSDE Sbjct: 648 SHVYSEVLHNDP----PSL-RPRWGSGHYVIPATGEPVERDAPFDPHYDLVTNYESDSDE 702 Query: 708 GSLFTLSSISSESARSKTEEAVPDE----ESLQDESSGASKDNVTAVDSLEENVTFQTI 762 GSLFTLSS SE RS EE E + L + G KD+VT+ +S+E+ +T Q I Sbict: 703 GSLFTLSSEGSEDTRSLAEEQASVENDGTSQPLPSRNLGEYKDSVTSAESVED-LTSQRI 761 Query: 763 PGKCKNQEDPFEKPLISAPDSGMYKTHLENASDTDRSEGLSPWPRSPGNSPLGDEFPGMF 822 P KC+ QE LIS PDS + +T+ EN S + E S WP+ PG+ Sbjct: 762 PEKCEAQEAHLRNTLISGPDSCVCETNQENDSSSLDPENRSTWPQLPGHKLSHHETLGTY 821

Query: 823 TYDYDTALQSKAAEWHCSLRDLEFSNVDVLQQTPPCSAEVPSDPDKAA 870 D QS+A +WH SLRDLE NVD +PP S E S P+ A

Sbjct: 822 G---DIEPQSEAVDWHYSLRDLESPNVD-SSPSPPYSDEDLSGPEDRA 865

Figure 3: Domain Professor results for INSP179

Query: INSP179.pp



E-values and regions on this page use repsilon and are approximate - hit "Aln" for Blastpgp evalues and alignments

| 11 | SCOP von<br>Willebrand | 82% | c.10.2.7 (A:) von Willebrand<br>factor binding domain of<br>glycoprotein Ib alpha {Human<br>(Homo sapiens)}   | d1gwba_          | 35-271  | 4-221         | -/-               | <u>2</u> /1.4e-25  |
|----|------------------------|-----|---|------------------|---------|---------------|-------------------|--------------------|
| 2  | N . 26 - 37 - 1        | 82% | 3:80:10:10 Alpha Beta;<br>rforseshoe; Ribonuclease<br>Inhibitor;  | Mr. 93           | 35-271  | 4-221         | - (               | <u>2</u> / 1.4e-25 |
| 3  | IPL<br>Whole_length    |     | Ferrodoxin-like / Adenylyl and guanylyl cyclase catalytic domain Ascomycotal and Fungal ACs, most of the full length alignment. Have added hCP46367.1 and hCP46942.1 to the alignment for this profile. | <u>IPL002890</u> | 40-255  | 1060-<br>1254 | <u>1</u> /1.4e-15 | -/-                |
| 4  | CDD COG4886            | 33% | Leucine-rich repeat (LRR) protein<br>[Function unknown]   | COG4886.         | 90-238  | 98-228        | 1/5.3e-14         | /w.                |
| 5  | SPAN DOMAIN            | 31% | DOMAIN EXTRACELLULAR<br>(POTENTIAL)<br>sp LGR5_HUMAN O75473 1-<br>561   | <u>075473</u>    | 45-231  | 51-222        | <u>1</u> /3.0e-11 | -/-                |
| 6  | SPAN DOMAIN            | 81% | DOMAIN ASP/SER-RICH<br>sp SR40_YEAST P32583 25-314  | <u>P32583</u>    | 549-797 | 15-248        | 1/9.0e-05         | -/-                |

Figure 4: Signal peptide prediction (SignalP V2.0) for INSP179 polypeptide sequence (SEQ ID NO: 10).

>INSP179.pp SignalP-NN prediction (euk networks): INSP179.pp C score 1.0 S score Y score 0.8 0.6 0.4 0.2 0.0 MKNLYFRV I T IV IGLYFTG IMTNASRKSN I LFNSECOMNEY I LTNCSFTGKCD I PVD I SQTAATVDVSF 0 10 20 30 40 50 60 70 Position >INSP179.pp length = 70Position Value Cutoff signal peptide? # Measure max. C 0.179 0.33 25 max. Y 25 0.318 0.32 NO 0.933 0.82 YES max. S 9 0.697 0.47 YES 1 - 24mean S # Most likely cleavage site between pos. 24 and 25: TNA-SR SignalP-HMM prediction (euk models): INSP179.pp Cleavage prob. 1.0 n-region prob. h-region prob. c-region prob. 0.8 0.6 0.4 0.2 0.0 MKNLYFRVIT IV IGLYFTG IMTNASRKSN I LFNSECOMNEY I LTNCSFTGKCD I PVD I SQTAATVDVSFN

>INSP179.pp
Prediction:

0

Prediction: Signal anchor

Signal peptide probability: 0.065 Signal anchor probability: 0.778

10

Max cleavage site probability: 0.031 between pos. 24 and 25

Position

50

60

70

20

Figure 5: INSP179 DNA and polypeptide sequence

| 1<br>61 | ttccgaaatc<br>attgtgaata | ttaaattgaa<br>agtcaattca | aattaaattt<br>ttttcctttt | gctgcttatt<br>tccaggcatt | gctgttagtt<br>atgaaaaacc<br>m k n | tttatatatt<br>tctatttcag<br>l y f |
|---------|--------------------------|--------------------------|--------------------------|--------------------------|-----------------------------------|-----------------------------------|
|         |                          |                          |                          |                          | INSP                              | 179-CP1                           |
| 121     | agtcattacc               | atagttatag<br>i v i      | gtctttattt<br>g l y      | tactggaata<br>f t g i    | atgacaaatg<br>m t n               | catcaagaaa<br>a s r               |
| 181     | aagcaatatt<br>k s n i    | ttattcaatt<br>1 f n      | ctgaatgcca<br>s e c      | atggaatgaa<br>q w n e    | tatattctga<br>y i l               | caaattgttc<br>t n c               |
| 241     | ttttaccgga<br>s f t g    | aagtgtgata<br>k c d      | tacctgtgga<br>i p v      | catatcacag<br>d i s q    | acagcagcca<br>t a a               | ctgtggatgt<br>t v d               |
| 301     | aagtttcaat<br>v s f n    | ttctttagag<br>f f r      | ttctcttaca<br>v l l      | gtctcacacg<br>q s h t    | aaaaaagaag<br>k k e               |                                   |
| 361     |                          | gacctcagta<br>d l s      |                          |                          | accttaagcc<br>t l s               |                                   |
| 421     |                          |                          | taaacctcag<br>l n l      |                          | atccactccc<br>i h s               | tctcattgga<br>l s l               |
| 481     | _                        | cctaagtcct<br>p k s      |                          | acgccacaga<br>k r h r    | agcagcttca<br>s s f               | gaaacaggtt<br>r n r               |
| 541     |                          |                          |                          |                          | agtgacactc<br>s d t               |                                   |
| 601     | gtggaaactg<br>l w k l    | aagtcattgc<br>k s l      | agagtttgga<br>q s l      | tctgtcattc<br>d l s f    | aatgggatat<br>n g i               | tgcaaatagg<br>l q i               |
| 661     | gtggtctgat<br>g w s d    |                          |                          | ggagaatctc<br>l e n l    | tgtttaaaga<br>c l k               | gcaacaagat<br>s n k               |
| 721     |                          | ccccacaag<br>p p q       |                          | cctcaaaaaa<br>d l k k    |                                   | tagaccttag<br>ì d l               |
| 781     |                          |                          | tcctaccaat<br>i l p      |                          | gctctagaat<br>a l e               | ttccccatct<br>f p h               |
| 841     | agtggttgac<br>l v v d    | ttggctgata<br>l a d      | ataactggca<br>n n w      | gtgtgatgat<br>q c d d    | agtgtggcag<br>s v a               | tctttcaaaa<br>v f q               |
| 901     | ttttatttct<br>n f i s    | gaatcctgga<br>e s w      | ggaaaaagtg<br>r k k      | gaatgtcatt<br>w n v i    | tgcaacaggt<br>c n r               | ctatagggag<br>s i g               |
| 961     | tgaggaggcc<br>s e e a    | aacgggggca<br>n g g      | ctccccagag<br>t p q      | caggatttcc<br>s r i s    | agggaaaccc<br>r e t               | gccttcctcc<br>r l p               |
| 1021    |                          |                          |                          |                          | gcagagaggc<br>a e r               | cccagggagg                        |
| 1081    | aaggcacacg<br>g r h t    | ggcatttcta<br>g i s      | ctctggggaa<br>t l g      | gaaggcaaag<br>k k a k    | gccggctctg<br>a g s               | gtctcaggaa<br>g l r               |

| 1141 | gaagcagaga<br>k k q r | cggctgccaa<br>r l p |                     | r s t r               | q A d               | ctgccggcaa<br>a a g |
|------|-----------------------|---------------------|---------------------|-----------------------|---------------------|---------------------|
|      |                       |                     |                     | INSP179-CP            | 2                   |                     |
| 1201 | aaaagaggac            | gctccccagg          | acctggctct          | ggcggtgtgc            | ctgtcagtgt          | tcatcacatt          |
|      | k k e d               | a p q               | d l a               | l a v c               | l s v               | f i t               |
| 1261 | ccttgtcgcc            | ttcagcctgg          | gggctttcac          | aaggccttat            | gttgacagac          | tgtggcaaaa          |
|      | f l v a               | f s l               | g a f               | t r p y               | v d r               | l w q               |
| 1321 | aaagtgccag            | agcaaaagcc          | ctggcctgga          | caacgcgtat            | tcaaacgagg          | gcttctacga          |
|      | k k c q               | s k s               | p g l               | d n a y               | s n e               | g f y               |
| 1381 | tgacatggaa<br>d d m e | gctgcggggc<br>a a g | acacaccaca<br>h t p | cccagagacc<br>h p e t | catctgcgcc<br>h 1 r |                     |
| 1441 | tcatctaagc            | ctctacgaga          | accagacccc          | tttctgggtg            | acacagccac          | acccacacgc          |
|      | p h l s               | l y e               | n q t               | p f w v               | t q p               | h p h               |
| 1501 | caccgtaatt            | cctgatagaa          | ctctgggaag          | gagcagaaag            | gatcctggca          | gttcgcagag          |
|      | a t v i               | p d r               | t l g               | r s r k               | d p g               | s s q               |
| 1561 | cccaggacag<br>s p g q | tgcggggaca<br>c g d | acaccggggc<br>n t g | aggaagtgga<br>a g s g |                     | cagtctattc<br>a v y |
| 1621 | cattctccag<br>s i l q | agacatccac<br>r h p | atgccggtaa<br>h a g | ccgtgaacta<br>n r e l | atgtcagcag<br>m s a |                     |
| 1681 | catccatagg            | aatgatattc          | tcggagaatg          | gacttatgaa            | actgtggccc          | aggaagagcc          |
|      | h i h r               | n d i               | l g e               | w t y e               | t v a               | q e e               |
| 1741 | tctcagtgca<br>p 1 s a | cattcagtgg<br>h s v |                     | tgtagctggc<br>s v a g |                     | ctgtctctgg<br>a v s |
| 1801 | ctcaagccgt            | tatgattcca          | atgaattaga          | cccttccctc            | tccggagaaa          | taacagcttc          |
|      | g s s r               | y d s               | n e l               | d p s l               | s g e               | i t a               |
| 1861 | cctctgtaaa            | atgctaacac          | atgcagaagc          | acagaggact            | ggagatagta          | aggaaagagg          |
|      | s l c k               | m l t               | h a e               | a q r t               | g d s               | k e r               |
| 1921 | gggcactgaa            | cagtcacttt          | gggactcgca          | gatggaattt            | tctaaggaaa          | ggcaagtgag          |
|      | g g t e               | q s l               | w d s               | q m e f               | s k e               | r q v               |
| 1981 | ttcatccatt            | gatttgctga          | gcatacagca          | gccaaggctg            | tccggggcaa          | gggctgagga          |
|      | s s s i               | d l l               | s i q               | q p r l               | s g a               | r a e               |
| 2041 | agcgctttca            | gcccactaca          | gcgaggttcc          | atacggtgac            | ccaagagaca          | caggcccatc          |
|      | e a l s               | a h y               | s e v               | p y g d               | p r d               | t g p               |
| 2101 | agtctttcct            | ccaagatggg          | acagtggcct          | ggatgtcact            | cctgctaaca          | aggaaccagt          |
|      | s v f p               | prw                 | d s g               | l d v t               | p a n               | k e p               |
| 2161 | gcagaaatco            | actccttctg          | acacttgctg          | tgagttggag            | agtgactgtg          | actctgatga          |
|      | v q k s               | t p s               | d t c               | c e l e               | s d c               | d s d               |
| 2221 | ggggtctctg            | ttcactctga          | gctccataag          | ttcagagagt            | gcaaggagca          | agactgaaga          |
|      | e g s l               | f t l               | s s i               | s s e s               | a r s               | k t e               |
| 2281 | ggcagtgcct            | gatgaggagt          | ccctgcagga          | . cgagagctca          | ggggcaagca          | aggacaatgt          |
|      | e a v p               | d e e               | s l q               | d e s s               | g a s               | k d n               |

| 2341 | gacggctgta | gacagtcttg | aggaaaatgt | taccttccaa | acaattccag | ggaaatgcaa |
|------|------------|------------|------------|------------|------------|------------|
|      | vtav       | d s l      | e e n      | vtfq       | tip        | g k c      |
| 2401 | gaatcaagaa | gatccctttg | aaaaacctct | catttctgct | ccagactctg | gcatgtacaa |
|      | k n q e    | d p f      | e k p      | lisa       | p d s      | g m y      |
| 2461 | gactcatctg | gaaaatgcct | ctgacactga | tagatctgag | ggcctgtcac | cctggcccag |
|      | k t h 1    | e n a      | s d t      | d r s e    | g l s      | p w p      |
| 2521 | gtcaccaggg | aatagtccct | taggggatga | gtttccgggc | atgttcactt | atgattatga |
|      | r s p g    | n s p      | l g d      | e f p g    | m f t      | y d y      |
| 2581 | cacagetett | caatccaagg | cagcagaatg | gcattgctca | cttagagact | tagaattttc |
|      | dtal       | q s k      | a a e      | whcs       | l r d      | l e f      |
| 2641 | aaatgtggac | gttttacagc | aaacaccacc | atgttctgct | gaagttccct | cagatectga |
|      | s n v d    | v l q      | q t p      | pcsa       | e v p      | s d p      |
| 2701 | taaggctgcc | ttccatgaaa | gagactcaga | cattttaaaa | taagaatctt | tcattaagga |
|      | dkaa       | f h e      | r d s      | dilk       |            |            |
| 2761 | aatattcaca | gctctaaaca | atattctttt | aaagatcatg | gcaggggaaa | actaaagcct |
| 2821 |            | atcctgaagg |            |            | aagtggacac | tgatgcaaat |
| 2881 | gagggttttg | tgggcccact | cgagggcgac | 9466       |            |            |

Position and sense of PCR primers

Figure 6: INSP179-EC DNA and polypeptide sequence

| 1   | atgaaaaacc | tctatttcag  | agtcattacc  | atagttatag | gtctttattt | tactggaata |
|-----|------------|-------------|-------------|------------|------------|------------|
|     | m k n      | l y f       |             | i v i      | g l y      | ftgi       |
|     |            | INSP179-CP1 |             |            |            |            |
| 61  | atgacaaatg | catcaagaaa  | aagcaatatt  | ttattcaatt | ctgaatgcca | atggaatgaa |
|     | _          | a s r       | k s n i     | l f n      | s e c      | q w n e    |
| 121 | tatattctga | caaattgttc  | ttttaccgga  | aagtgtgata | tacctgtgga | catatcacag |
|     | y i ĺ      | t n c       | s f t g     | k c d      | i p v      | d i s q    |
| 181 | acagcagcca | ctgtggatgt  | aagtttcaat  | ttctttagag | ttctcttaca | gtctcacacg |
|     | t a a      | t v d       | v s f n     | f f r      | v 1 1      | q s h t    |
| 241 | aaaaaagaag | agtggaaaat  | aaaacatctg  | gacctcagta | acaatctcat | atcaaaaata |
|     | k k e      | e w k       | i k h l     | d l s      | n n 1      | iski       |
| 301 | accttaagcc | cttttgcata  | tttacatgct  | ttggaagtgt | taaacctcag | caacaatgcc |
|     |            |             |             |            | l n l      |            |
| 361 | atocactoco | teteatteea  | tctactcact  | cctaadtcct | catgggtgaa | acccacaca  |
| 301 |            | l s l       |             |            |            | k r h r    |
| 421 | agcagcttca | gaaacaggtt  | tccattgctg  | aaggtgctca | ttcttcaaag | aaataaactc |
|     | s s f      | r n r       | f p l l     | k v l      | i l q      | r n k l    |
| 481 | agtgacactc | ccaagggact  | gtggaaactg  | aagtcattgc | agagtttgga | tctgtcattc |
|     | s d t      | p k g       | l w k l     | k s l      | q s l      | d l s f    |
| 541 | aatgggatat | tgcaaatagg  | gtggtctgat  | tttcacaact | gcctgcaact | ggagaatctc |
|     | n g i      | l q i       | g w s d     | f h n      | c l q      | l e n l    |
| 601 | tatttaaaga | gcaacaagat  | attcaaaatt  | ccccacaaq  | ccttcaagga | cctcaaaaaa |
| 001 | c l k      | s n k       | i f k i     | p q q      |            | d 1 k k    |
| 661 | ttacaddtca | tagaccttag  | caacaatgct  | ctgattacca | tcctaccaat | gatgatcata |
| 001 |            |             |             |            | i 1 p      |            |
| 721 | gctctagaat | ttccccatct  | agtggttgac  | ttggctgata | ataactggca | gtgtgatgat |
|     |            |             |             |            | n n w      |            |
| 781 | agtgtggcag | tctttcaaaa  | ttttatttct  | gaatcctgga | ggaaaaagtg | gaatgtcatt |
|     |            | v f q       | n f i s     |            | r k k      | wnvi       |
| 841 | tacaacaaat | ctatadddad  | taaaaaaaacc | aacgggggga | ctcccagag  | caggatttcc |
| 041 | c n r      | s i g       | s e e a     | n g g      | t p q      | s r i s    |
| 901 | agggaaaccc | accttectee  | cattcatcto  | catcgcatga | aaagcctcat | aaggagcaaa |
|     | r e t      |             | p i h l     |            | k s l      | i r s k    |
| 961 | acaasasaca | cccsaaasaa  | aaddcacacd  | agcatttcta | ctctggggaa | daaddcaaad |
| 201 | a e r      | p q g       | g r h t     |            | t 1 g      | k k a k    |

Position and sense of PCR primers ----

## Figure 7: INSP179-EC-SV1 DNA and polypeptide sequence

| 1   | atgaaaaacc<br>m k n | tctatttcag<br>l y f | agtcattacc<br>r v i t | atagttatag<br>i v i | gtctttattt<br>g l y | tactggaata<br>f t g i |
|-----|---------------------|---------------------|-----------------------|---------------------|---------------------|-----------------------|
|     |                     | INSP179-CP1         |                       |                     |                     |                       |
| 61  | atgacaaatg<br>m t n | catcaagaaa<br>a s r | aagcaatatt<br>k s n i | ttattcaatt<br>l f n | ctgaatgcca<br>s e c | atggaatgaa<br>q w n e |
| 121 | tatattctga<br>y i l | caaattgttc<br>t n c | ttttaccgga<br>s f t g | aagtgtgata<br>k c d | tacctgtgga<br>i p v |                       |
| 181 |                     | ctgtggatgt<br>t v d | aagtttcaat<br>v s f n | ttctttagag<br>f f r |                     |                       |
| 241 |                     | agtggaaaat<br>e w k | aaaacatctg<br>i k h l |                     | acaatctcat<br>n n l | atcaaaaata<br>i s k i |
| 301 | accttaagcc<br>t l s | cttttgcata<br>p f a | tttacatgct<br>y l h a | ttggaagtgt<br>l e v | taaacctcag<br>l n l | caacaatgcc<br>s n n a |
| 361 | atccactccc<br>i h s |                     | tctactcagt<br>d l l s |                     |                     |                       |
| 421 | agcagcttca<br>s s f | gaaacaggtt<br>r n r | tccattgctg<br>f p l l | aaggtgctca<br>k v l | ttcttcaaag<br>i l q | aaataaactc<br>r n k l |
| 481 | agtgacactc<br>s d t |                     | tgaggaggcc<br>s e e a | aacgggggca<br>n g g |                     | caggatttcc<br>s r i s |
| 541 | agggaaaccc<br>r e t | gccttcctcc<br>r l p | cattcatctg<br>p i h l | catcgcatga<br>h r m | aaagcctcat<br>k s l | aaggagcaaa<br>i r s k |
| 601 | gcagagaggc<br>a e r | cccagggagg<br>p q g | aaggcacacg<br>g r h t |                     | ctctggggaa<br>t l g |                       |
| 661 | gccggctctg<br>a g s | gtctcaggaa<br>g l r | gaagcagaga<br>k k q r | cggctgccaa<br>r l p | ggagtgttag<br>r s v | aagcacccgc<br>r s t r |
| 721 | gatgtgcag<br>d v q  |                     |                       |                     | IN                  | SP179-CP2             |

Figure 8: Alignment of INSP179-EC and INSP179-EC-SV1 nucleotide sequences

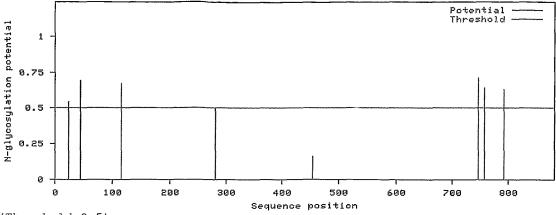
| 179EC-SV1              | ATGAAAAACCTCTATTTCAGAGTCATTACCATAGTTATAGGTCTTTATTTTACTGGAATA  |
|------------------------|---|
| INSP179EC              | ATGAAAAACCTCTATTTCAGAGTCATTACCATAGTTATAGGTCTTTATTTTACTGGAATA  |
| 179EC-SV1              | ATGACAAATGCATCAAGAAAAAGCAATATTTTATTCAATTCTGAATGCCAATGGAATGAA  |
| INSP179EC              | ATGACAAATGCATCAAGAAAAAGCAATATTTTATTCAATTCTGAATGCCAATGGAATGAA  |
| 179EC-SV1              | TATATTCTGACAAATTGTTCTTTTACCGGAAAGTGTGATATACCTGTGGACATATCACAG  |
| INSP179EC              | TATATTCTGACAAATTGTTCTTTTACCGGAAAGTGTGATATACCTGTGGACATATCACAG  |
| 179EC-SV1              | ACAGCAGCCACTGTGGATGTAAGTTTCAATTTCTTTAGAGTTCTCTTACAGTCTCACACG  |
| INSP179EC              | ACAGCAGCCACTGTGGATGTAAGTTTCAATTTCTTTAGAGTTCTCTTACAGTCTCACACG  |
| 179EC-SV1              | AAAAAAGAAGAGTGGAAAATAAAACATCTGGACCTCAGTAACAATCTCATATCAAAAATA  |
| INSP179EC              | AAAAAAGAAGAGTGGAAAATAAAACATCTGGACCTCAGTAACAATCTCATATCAAAAATA  |
| 179EC-SV1              | ACCTTAAGCCCTTTTGCATATTTACATGCTTTGGAAGTGTTAAACCTCAGCAACAATGCC  |
| INSP179EC              | ACCTTAAGCCCTTTTGCATATTTACATGCTTTGGAAGTGTTAAACCTCAGCAACAATGCC  |
| 179EC-SV1              | ATCCACTCCCTCTCATTGGATCTACTCAGTCCTAAGTCCTCATGGGTGAAACGCCACAGA  |
| INSP179EC              | ATCCACTCCCTCTCATTGGATCTACTCAGTCCTAAGTCCTCATGGGTGAAACGCCACAGA  |
| 179EC-SV1              | AGCAGCTTCAGAAACAGGTTTCCATTGCTGAAGGTGCTCATTCTTCAAAGAAATAAACTC  |
| INSP179EC              | AGCAGCTTCAGAAACAGGTTTCCATTGCTGAAGGTGCTCATTCTTCAAAGAAATAAACTC  |
| 179EC-SV1<br>INSP179EC | AGTGACACTCCCAAGGGAGTGACACTCCCCAAGGGACTGTGGAAACTGAAGTCATTGCAGAGTTTGGATCTGTCATTC  |
| 179EC-SV1<br>INSP179EC | AATGGGATATTGCAAATAGGGTGGTCTGATTTTCACAACTGCCTGC  |
| 179EC-SV1<br>INSP179EC | TGTTTAAAGAGCAACAAGATATTCAAAAATTCCCCCACAAGCCTTCAAGGACCTCAAAAAA   |
| 179EC-SV1<br>INSP179EC | TTACAGGTCATAGACCTTAGCAACAATGCTCTGATTACCATCCTACCAATGATGATCATA  |
| 179EC-SV1<br>INSP179EC | GCTCTAGAATTTCCCCATCTAGTGGTTGACTTGGCTGATAATAACTGGCAGTGTGATGAT  |
| 179EC-SV1<br>INSP179EC | AGTGTGGCAGTCTTTCAAAATTTTATTTCTGAATCCTGGAGGAAAAAGTGGAATGTCATT  |
| 179EC-SV1<br>INSP179EC | GAGTGAGGAGGCCAACGGGGCACTCCCCAGAGCAGGATTTCC TGCAACAGGTCTATAGGGAGTGAGGAGGCCAACGGGGGCACTCCCCAGAGCAGGATTTCC                   |
| 179EC-SV1              | AGGGAAACCCGCCTTCCTCCCATTCATCTGCATCGCATGAAAAGCCTCATAAGGAGCAAA  |
| INSP179EC              | AGGGAAACCCGCCTTCCTCCCATTCATCTGCATCGCATGAAAAGCCTCATAAGGAGCAAA  |
| 179EC-SV1<br>INSP179EC | GCAGAGAGGCCCCAGGGAGGAAGGCACACGGGCATTTCTACTCTGGGGAAGAAGGCAAAGGCAGAGAGGGCCCCAGGGAGGAAGGCACACGGGCATTTCTACTCTGGGGAAGAAGGCAAAG |
| 179EC-SV1              | GCCGGCTCTGGTCTCAGGAAGAAGCAGAGACGGCTGCCAAGGAGTGTTAGAAGCACCCGC  |
| INSP179EC              | GCCGGCTCTGGTCTCAGGAAGAAGCAGAGACGGCTGCCAAGGAGTGTTAGAAGCACCCGC  |
| 179EC-SV1              | GATGTGCAG   |
| INSP179EC              | GATGTGCAG   |

## Figure 9: Alignment of predicted INSP179 and the cloned INSP179-EC and INSP179-EC-SV1 polypeptide sequences

| INSP179-EC_PREDICTION INSP179-EC_CLONED INSP179-EC-SV1_CLONED | MKNLYFRVITIVIGLYFTGIMTNASRKSNILFNSECQWNEYILTNCSFTG<br>MKNLYFRVITIVIGLYFTGIMTNASRKSNILFNSECQWNEYILTNCSFTG<br>MKNLYFRVITIVIGLYFTGIMTNASRKSNILFNSECQWNEYILTNCSFTG<br>************************************ |
|---|--|
| INSP179-EC_PREDICTION INSP179-EC_CLONED INSP179-EC-SV1_CLONED | KCDIPVDISQTAATVDVSFNFFRVLLQSHTKKEEWKIKHLDLSNNLISKI<br>KCDIPVDISQTAATVDVSFNFFRVLLQSHTKKEEWKIKHLDLSNNLISKI<br>KCDIPVDISQTAATVDVSFNFFRVLLQSHTKKEEWKIKHLDLSNNLISKI<br>***********************************  |
| INSP179-EC_PREDICTION INSP179-EC_CLONED INSP179-EC-SV1_CLONED | TLSPFAYLHALEVLNLSNNAIHSLSLDLLSPKSSWVKRHRSSFRNRFPLL<br>TLSPFAYLHALEVLNLSNNAIHSLSLDLLSPKSSWVKRHRSSFRNRFPLL<br>TLSPFAYLHALEVLNLSNNAIHSLSLDLLSPKSSWVKRHRSSFRNRFPLL<br>***********************************  |
| INSP179-EC_PREDICTION INSP179-EC_CLONED INSP179-EC-SV1_CLONED | KVLILQRNKLSDTPKGLWKLKSLQSLDLSFNGILQIGWSDFHNCLQLENL<br>KVLILQRNKLSDTPKGLWKLKSLQSLDLSFNGILQIGWSDFHNCLQLENL<br>KVLILQRNKLSDTPK  |
| INSP179-EC_PREDICTION INSP179-EC_CLONED INSP179-EC-SV1_CLONED | CLKSNKIFKIPPQAFKDLKKLQVIDLSNNALITILPMMIIALEFPHLVVD<br>CLKSNKIFKIPPQAFKDLKKLQVIDLSNNALITILPMMIIALEFPHLVVD   |
| INSP179-EC_PREDICTION INSP179-EC_CLONED INSP179-EC-SV1_CLONED | LADNNWQCDDSVAVFQNFISESWRKKWNVICNRSIGSEEANGGTPQSRIS LADNNWQCDDSVAVFQNFISESWRKKWNVICNRSIGSEEANGGTPQSRISGSEEANGGTPQSRIS ***********************************   |
| INSP179-EC_PREDICTION INSP179-EC_CLONED INSP179-EC-SV1_CLONED | RETRLPPIHLHRMKSLIRSKAERPQGGRHTGISTLGKKAKAGSGLRKKQR<br>RETRLPPIHLHRMKSLIRSKAERPQGGRHTGISTLGKKAKAGSGLRKKQR<br>RETRLPPIHLHRMKSLIRSKAERPQGGRHTGISTLGKKAKAGSGLRKKQR<br>**********************************   |
| INSP179-EC_PREDICTION INSP179-EC_CLONED INSP179-EC-SV1_CLONED | RLPRSVRSTRDVQ<br>RLPRSVRSTRDVQ<br>RLPRSVRSTRDVQ<br>*********   |

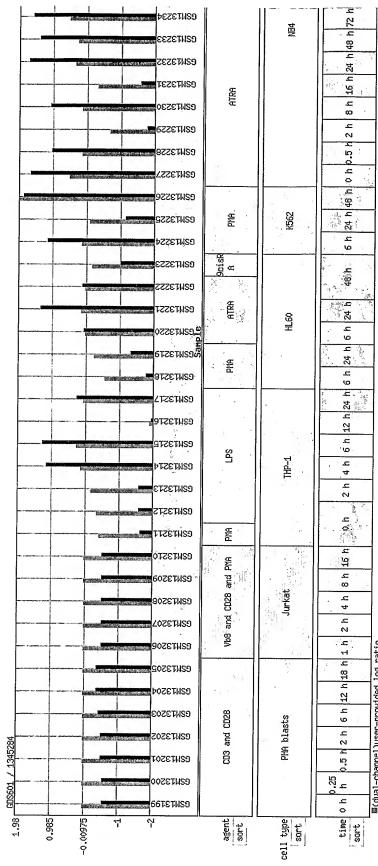
Figure 10: Predicted INSP179 glycosylation sites

NetNGlyc 1.0: predicted N-glycosylation sites in Sequence



(Threshold=0.5)

| SeqName  | Position | Potential | Score |  |
|----------|----------|-----------|-------|--|
|          |          |           |       |  |
| Sequence | 23 NASR  | 0.5420    | (6/9) |  |
| Sequence | 45 NCSF  | 0.6932    | (9/9) |  |
| Sequence | 115 NLSN | 0.6702    | (9/9) |  |
| Sequence | 282 NRSI | 0.4953    | (4/9) |  |
| Sequence | 454 NQTP | 0.1590    | (9/9) |  |
| Sequence | 746 NVTA | 0.7102    | (9/9) |  |
| Sequence | 756 NVTF | 0.6390    | (9/9) |  |
| Sequence | 792 NASD | 0.6288    | (6/9) |  |



⊠(dual-channel)user-provided log ratio ■percentile ranked and binned value of a spot compared to all other spots within that sample